

The diagnostic methods in hematology

Poltava State Medical University
The Internal Medicine №1 Department

2021

Plan of the lecture

I. Hematopoiesis

1. Hematopoietic organs

II. General Outlines. The main syndromes

1. Anemic

2. Hemorrhagic

3. Hyperplastic syndrome

4. Ulcerative-necrotic syndrome

5. Lymphadenopathy

III. Laboratory diagnostics

1. General blood analysis

2. Blood chemistry

3. Spinal puncture + counting myelogram

4. Morphological examination of bone marrow cells

5. Cytochemical study of bone marrow

6. Immunological examination of bone marrow and peripheral blood cells

7. Cytogenetic examination

8. Molecular study

9. Biopsy of target lymph node + immunohistochemical examination

I. Hematopoiesis

The volume of blood in adults is 5-5.5 liters - 1/13 of weight;
55-60% - plasma; 40-45% - cellular elements

The source of hematopoiesis - PSC - properties:

- Polypotency - ability to multilinear differentiation
- Ability to self-maintain - consistency of the pool

Hemopoiesis is a process that consists of successive divisions and differentiations, resulting in the formation of mature blood cells:

Erythrocytes - transfer of oxygen

Platelets - participation in hemostasis

Basophilic, eosinophilic, neutrophilic granulocytes, monocytes / macrophages - cells of natural nonspecific immunity

B- and T-lymphocytes are cells of humoral and cellular immunity

Central hematopoietic organs

Bone marrow: ripening of myelopoiesis elements,

I stage of maturation of B-lymphocytes

- in flat bones (skull, sternum, ribs, vertebrae, pelvic bones)**
- in the tubular bones epiphyses (hip, shoulder, shin)**

Thymus - T-lymphocytes maturation

Peripheral hematopoietic organs

Spleen

Lymphatic nodes

**Mucosal associated lymphoid tissue
(MALT)**

Peripheral blood (PSC)

Diseases of Blood and Blood forming organs

A - Benign Hematology

- * 1- Bleeding Disorders**
- * 2- Thrombophilia**
- * 3- Anemia**
- * 4- WBC disorders/non-malignant**
- * 5- BM disorders/ non-malignant (AA)**
- * 6- Secondary to other diseases (reactive, pancytopenia... ...etc)**

Diseases of Blood and Blood forming organs

B - Malignant Hematology

- * **1- Leukemias: acute/chronic**
- * **2- Lymphomas: NHL/ HL**
- * **3- Plasma cell descrecias: MM**
- * **4- Myeloproliferative neoplasms (MPN)**

II. General Outlines

- * **Symptoms and signs may not distinguish between various etiologies and syndromes**
- * **Severity of symptoms may vary according to many factors**
- * **Symptoms and signs may be part of the syndrome causing a hematological disease**
- * **Symptoms and signs may overlap with non-hematological disease**

Signs in Hematology

- * **Frequently non-specific**
- * **May be characteristic**
- * **Combination of the abnormalities causing the symptoms**
- * **May be very apparent in advanced disease or very subtle in early disease**
- * **Careful examination is needed**
- * **Changing signs require caution and repeated examination**

Clinical examination of patients

The main syndromes of the hematopoiesis system diseases :

- **Anemic**
- **Hemorrhagic**
- **Intoxicating**
- **Tumor proliferation syndrome**
- **Infectious complications syndrome**

Features of complaints in hematological diseases

Common symptoms:

Unmotivated general weakness

Dyspnea with physical exertion

Headache

Anemic syndrome:

General weakness

Dizziness, headache

Tinnitus (noise at ears)

Dyspnea with usual physical exertion

II.1. Anemic syndrome - clinical manifestations depend on:

- 1. Degree of blood oxygen saturation ↓ - degree of hypoxia**
- 2. Degree of total blood volume ↓**
- 3. The time of development of the first two factors**
- 4. Manifestations of the underlying disease**
- 5. The ability of CVS and RS to compensate anemia**

Symptoms characteristic of different anemia types

- * **Dry skin, brittle nails, hair loss, perversion of taste (desire is chalk) - *sideropenic syndrome* - (IDA)**
- * **Yellowing of the skin, icteric sclera, darkening of urine - *hemolytic syndrome* - (B12 deficiency anemia, hemolytic anemia)**
- * ***Neurological Syndrome:***
Paresthesia, disorder of superficial sensitivity, sometimes pareses, memories ↓ - B12 deficiency anemia
- * **GI tract disorders: depends on the severity of the lesion**

Complaints

Hemorrhagic syndrome:

Bleeding gums,

Hemorrhages on the skin

Bleeding of different localizations

Intoxication syndrome(hemoblastosis, hemolytic crises, aplastic anemia):

↓ appetit, weight loss

↑ temperature

Infectious complications syndrome

↑ temperature caused by infection

Manifestations of angina, otitis, bronchitis, pneumonia

Complaints

- *Ulcerative-necrotic syndrome*

- *Tumor proliferation syndrome* (leukemia acute, chronic)

- ↑ lymph nodes,

- ↑ spleen, liver - heaviness in the right and left hypochondrium

- symptoms of internal organs compression (ERW)

- rashes of a purple color - leukemids

- ossalgia - pain in long tubular bones, sternum

- *Tumor lysis syndrome*

- ↑ temperature,

- ↓ blood pressure

- artralgia (secondary gout)

- Kidney function disorder (ARF)

Anamnestic data:

- **Duration of symptoms**
- **Symptoms set**
- **The nature of the disease is hereditary, acquired**
- **Features of food - vegetarianism**
- **Occupational hazards - the effect of ionizing radiation, high frequency currents, chemicals**
- **Presence of oncological diseases in the anamnesis - cytostatics treatment**
- **Use of medicines - NSAIDs, glucocorticoids, mercazolyl, etc.**

Concomitant diseases

Chronic diseases of internal organs:

liver,

kidney,

diffuse connective tissue diseases

Gynecological diseases - with IDA:

duration of the menstrual cycle

duration of menstruation

number of pregnancies and births

the presence of anemia in pregnancy

**presence of fibromyoma, endometriosis, hyperplasia of the
uterus, malignant neoplasms**

Anamnestic data

- **Gastrointestinal lesions - latent bleeding**
- **Bleeding from varicose veins of the esophagus, neoplasms of the esophagus**
- **Stomach ulcers and duodenal ulcers, anacid gastritis, neoplasm of the stomach**
- **Resection of the stomach**
- **NU**
- **Polyps of the large intestine**
- **Bleeding from the hemorrhoidal nodes**
- **Malignant colon formation**

Objective examination

Pale skin

Paleness of mucus membranes

Yellowness of the skin

"Blue sclera" symptom

"Geographical tongue"

"Lacquered tongue "



II.2. Manifestations of hemorrhagic syndrome:

Disorders of platelet hemostasis:

Bruises

Fine petechial rash

Disorders of the coagulation hemostasis:

Hematomas

Hemorrhages in large joints (hemarthrosis)

Hemorrhagic syndrome with AL









Рисунок 2. Постинъекционные экхимозы
у пациента с тромбоцитопенией



Рис. 3. Геморрагическая сыпь (петахи, экхимозы) в разных стадиях обратного развития у больного с симптоматической тромбоцитопенией (приобретенная апластическая анемия)

Haemarthrosis of the left knee in hemophilia



Lesion of joints with hemophilia



*The pink-violet skin seals up to 1-2 cm -leukemids –
skin lesions at AL*

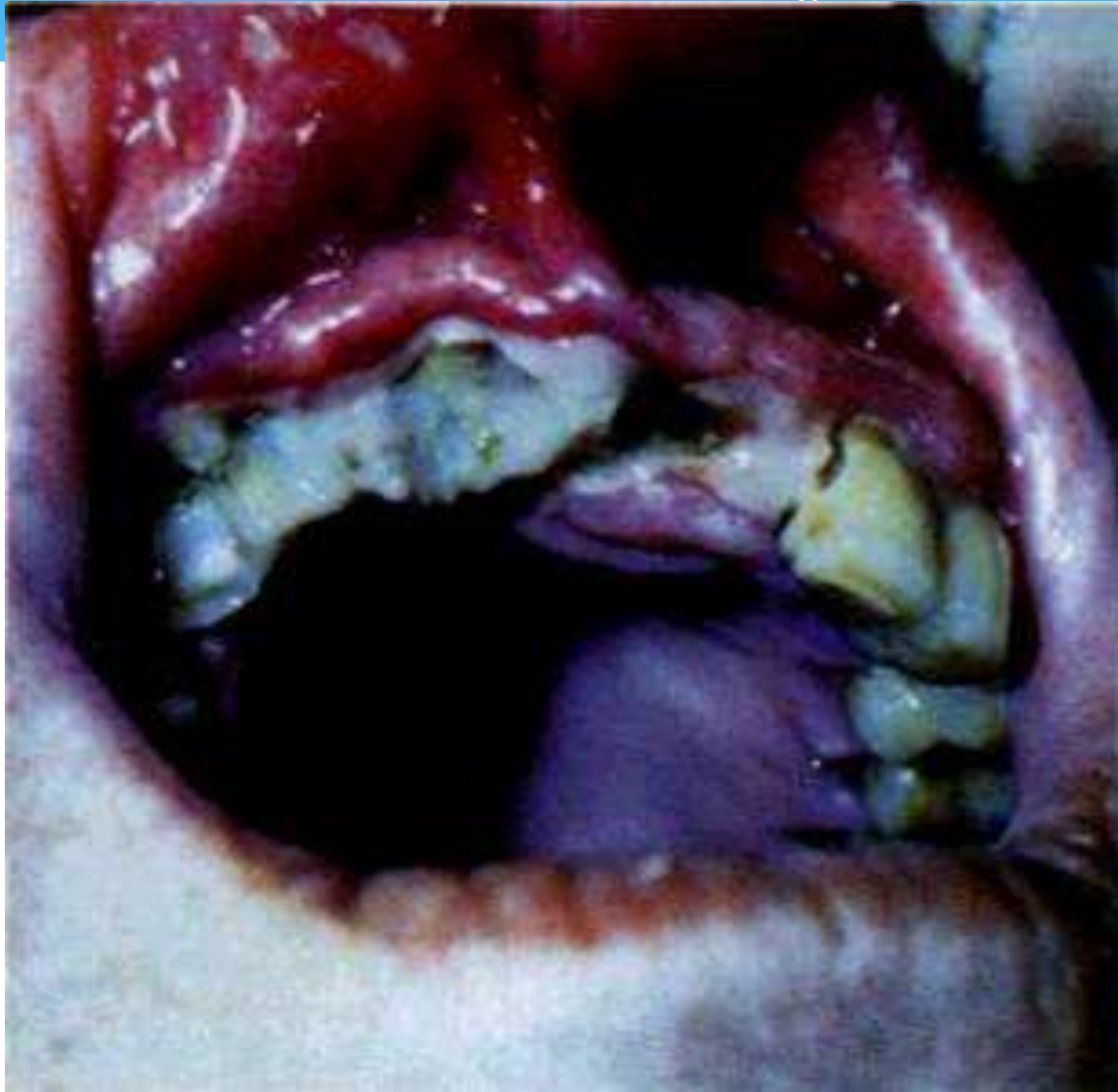




II.3. Hyperplastic syndrome with AL



II.4. Marginal necrosis of gums in patient with AL - ulcerative-necrotic syndrome



Objective examination

Palpation of peripheral lymph nodes:

- Dimensions
- Consistency
- Confusion, mobility
- Soreness
- Skin color over an enlarged lymph node

Determination of the size of the spleen - splenomegaly

Determination of liver size - hepatomegaly

Hyperplastic syndrome

II.5. Lymphadenopathy



Objective examination

Respiratory system changes :

*bronchitis, pneumonia, pleurisy, specific lung damage, symptoms of compression of the bronchial tree

Cardiovascular system changes :

*tachycardia, rhythm disturbances, weakening of the I tone on the apex, systolic murmur

Digestive system changes :

*erosive-ulcerative lesions of stomach and duodenum. Pain at palpation in the epigastric area of the abdomen

*enteropathy - painful palpation

Secondary Nephropathy - CRF

Neurological symptoms

- disorder of superficial/deep sensitivity, paresis

III. Laboratory diagnostics in hematology

- * **General blood analysis**
- * **Blood chemistry**
- * **Spernal puncture + counting myelogram**
- * **Morphological examination of bone marrow cells**
- * **Cytochemical study of bone marrow**
- * **Immunological examination of bone marrow and peripheral blood cells**
- * **Cytogenetic examination**
- * **Molecular study**
- * **Biopsy of target lymph node + immunohistochemical examination**

III.1. Hemogram indicators

Hemoglobin:

- * 120 - 140 g / l for women
- * 132 - 164 g / l for men

Hemoglobin concentration reduction (absolute):

- * with anemia of different genesis (the degree of anemia is determined by the degree of hemoglobin decrease)

Relative hemoglobin decrease:

during pregnancy due to an increase in the volume of plasma.

Increase in hemoglobin concentration:

- * with true polycythemia (PV)
- * symptomatic erythrocytosis.

From left to right: erythrocyte, thrombocyte, leucocyte.



Red blood cells

- * $4,5 - 5,1 \times 10^{12} / l$ - for men
- * $3,5 - 4,5 \times 10^{12} / l$ - for women

The number of erythrocytes in anemic syndrome is decreased

Absolute erythrocytosis

- * True polycythemia

Symptomatic erythrocytosis

- * Relative erythrocytosis - at ↓ volume of the liquid part of the blood (with prolonged vomiting, diarrhea, burns).

Diameter of erythrocytes: 7-8 μm

- * Microcytes - 5-6 μm
- * Macrocytes - more than 9 μm

Hematologic indices

The average volume of erythrocyte MCV is 80-95 fl,

** $MCV = 10 \times \text{hematocrit\%} / \text{erythrocyte count} \times 10^{12} / l$*

Mean hemoglobin content in one red cell MCH

** $MCH = \text{hemoglobin (g / l)} / \text{erythrocyte count} \times 10^{12} / l$*

** $MCH\ 26.7\text{-}33.3\ \text{pg}$*

*The average concentration of hemoglobin in the erythrocyte
MCHC*

** $MCHC = \text{hemoglobin (g / l)} \times 10 / \text{hematocrit (\%)}$*

** $MCHC\ 32\text{-}38\ \text{g / dL}$*

Color index

Characterizes the hemoglobin content in one erythrocyte

CI = hemoglobin × 3 / first three digits of red blood cells index

Norm 0.86 - 1.05

Hypochromic: IDA

Normochromic: hemolytic, aplastic anemia.

Hyperchromic: B-12 deficiency

Reticulocytes

- The norm is 0.8-1.2% or 2-15 ‰ of the total number of erythrocytes
- * Reticulocyte - young erythrocyte
- * The number of reticulocytes reflects the regenerative capacity of the bone marrow

Reticulocytosis is observed when:

True reticulocytosis (in both peripheral blood and BM)

- * Hemolytic anemia (hemolytic crisis)
- * Acute posthemorrhagic anemia

False reticulocytosis (isolated reticulocytosis in peripheral blood)

- * Metastasis of solid tumors in BM

Leukocytes

Normal values in the adult's hemogram

$4,0-9,0 \times 10^9 / l$

Physiological leukocytosis:

- * ↑ physical stress - myogenic leukocytosis**
- * Pregnancy (usually in the 2nd half),**
- * Emotional stress,**
- * 2-3 hours after meals, especially rich in protein - food leukocytosis**
- * After taking individual medications - glucocorticoids**

Pathological leukocytosis

- * **acute and chronic inflammatory processes, sepsis**
- * **infarction of different organs,**
- * **massive burns**
- * **intoxications,**
- * **hepatic, renal failure,**
- * **acute blood loss,**
- * **hemolytic crisis,**
- * **allergic diseases,**
- * **solid tumors, especially with tumor decay.**

Leukocytosis in the diseases of the blood system is characterized by:

- * Moderate - for most AL, non-Hodgkin's malignant lymphomas, true polycythemia;**
- * Expressed ($100-200 \times 10^9 / l$) - for chronic myelogenous leukemia (CML), less often for chronic lymphatic leukemia (CLL), sometimes for AL**

Leukopenia occurs when:

- * **Acute Leukemia, NLD**
- * **acute viral infections,**
- * **diffuse connective tissue diseases (SLE, Felty syndrome),**
- * **At a number of medications treatment (cytostatics, NSAIDs, sulfonamides, antibiotics).**

Leukocyte formula

*Counted on the basis of peripheral blood smears
morphological study*

* Stab neutrophils	1-6%	0,04-0,3
* Segmented neutrophils	47-72%	2,0-7,5
* Eosinophils	0,5-5%	0,02-0,3
* Basophils	0-1%	0-0,1
* Lymphocytes	19-37%	1,5-3,5
* Monocytes	2-10%	0,02-0,8

The value of the leukocyte formula

Shift of the leukocyte formula to the left - ↑ the number of immature neutrophils in the peripheral blood (myelocytes, metamyelocytes, stab neutrophils, sometimes with the appearance of blast cells):

**Acute and chronic inflammatory and infectious processes*

**Solid tumors*

**Sepsis*

**Chronic myeloproliferative diseases*

**AL*

The value of the leukocyte formula

Increase in the number of segmented neutrophils with hypersegmented nuclei - a shift of the leukocyte formula to the right:

- * Megaloblastic anemia*
- * Myelodysplastic syndrome*
- * Radiation sickness*
- * Solid tumors*
- * At the background of treatment with cytostatics*

The value of the leukocyte formula

Reduction in the number of neutrophils at less than $1.5 \times 10^9 / L$ - neutropenia - with:

- * Viral infections*
- * Chronic inflammatory processes*
- * Some hemoblastoses*
- * At the backdrop of radiation therapy*
- * At the backdrop of cytostatic therapy*
- * Agranulocytosis - neutrophils less than $0.5 \times 10^9 / L$*

Platelets

Normally, the number of platelets is $150-350 \times 10^9 / L$

Thrombocytopenia is noted in hematologic diseases:

- *immune and non-immune thrombocytopenia,*
- *B₁₂ deficiency anemia*
- *Aplastic anemia (AA)*
- *Hemolytic anemia*
- *Acute leukemia (AL),*
- *The later stages of CLL and CML,*
- *Multiple myeloma (MM),*
- *Myelodysplastic syndrome (MDS)*
- *Hairy cell leukemia (HCL),*
- *Non-Hodgkin's malignant lymphomas.*

Primary thrombocytosis:

- * **Idiopathic myelofibrosis (IMF)**
- * **Chronic myelogenous leukemia (CML)**
- * **Polycythemia Vera (PV)**
- * **Chronic myelomonocytic leukemia**
- * **Myelodysplastic syndrome (MDS):**
 - **with the 5q deletion**
 - **refractory anemia with ring sideroblasts and thrombocytosis**

Secondary thrombocytosis (short-term)

- * **Physical exertion**
- * **Acute infectious diseases**
- * **Therapy with vitamin B₁₂**
- * **Allergic reactions**
- * **Tissues injury**
- * **Myocardial infarction**
- * **Acute pancreatitis**
- * **Acute hemorrhage or hemolysis**
- * **Medication-induced (corticosteroids, adrenaline, vincristine, interleukin 1 β , epinephrine, transretinic acid)**

Secondary chronic thrombocytosis

- * **Iron Deficiency**
- * **Surgical and functional asplenia**
- * **Chronic infections**
- * **Rheumatological diseases**
- * **Systemic amyloidosis**
- * **Chronic inflammatory diseases**
- * **Celiac disease**
- * **Malignant neoplasms**
- * **Chronic kidney disease**

Erythrocytes sedimentation rate (ESR)

Normal 1-10 mm / h men; 2-15 mm / h women

Acceleration of ESR is noted :

- * Acute and chronic infectious and inflammatory processes,**
- * Immunosecretizing tumors (MM, Waldenstrom macroglobulinemia),**
- * Symptomatic paraproteinemia in the background of chronic lymphocytic leukemia (CLL) and non-Hodgkin's malignant lymphomas (NLL)**
- * Autoimmune diseases,**
- * Solid tumors.**

Reduction of ESR is observed at

- * PV, symptomatic erythrocytosis, viral hepatitis, mechanical jaundice.**

Міністерство охорони здоров'я України

Найменування закладу

КДЛ ПОКЛ

34

МЕДИЧНА ДОКУМЕНТАЦІЯ

Форма № 224/о

Затверджена наказом МОЗ України

04.01.2001 р. № 1

КЛІНІЧНИЙ АНАЛІЗ КРОВІ №

72186

Прізвище

Заклад

Відділення

H-100

Медична карта №

Клінічний діагноз (профогляд)

Anemia

Найменування показників

Результат

Гемоглобін

ч

ж

Еритроцити

ч

ж

Кольоровий показник

Ретикулоцити

Тромбоцити

Лейкоцити

Швидкість осідання еритроцитів (ШОЕ)

ч

ж

Нейтрофіли

Мієлоцити

Метамієлоцити

Паличкоядерні

Сегментоядерні

Еозинофіли

Базофіли

Лімфоцити

Моноцити

Плазматичні клітини

KDL POKL

Sysmex

XP-300

Operator

ID. 34

Date 24/07/2014

Time 12:13

Mode WB

WBC WL* $9.0 \times 10^9/L$ RBC $3.90 \times 10^{12}/L$

HGB - 65 g/L

HCT - 0.245

MCV - 62.8 fL

MCH - 16.7 pg

MCHC - 265 g/L

PLT AG* $475 \times 10^9/L$

WBC



100 200 300 [fL]

LYM% WL* 0.254

MXD% WL* 0.059

NEUT% WL* 0.687

LYM# WL* $2.3 \times 10^9/L$ MXD# WL* $0.5 \times 10^9/L$

Iron metabolism study

Serum iron (9.0 - 31.3 $\mu\text{mol} / \text{L}$)

The total iron-binding capacity of blood(50-65 $\mu\text{mol} / \text{l}$)

Serum ferritin (20-160 $\mu\text{g} / \text{L}$)

Osmotic resistance of erythrocytes:

- * **0,50-0,45% the beginning of hemolysis,**
- * **0,35-0,40 - maximum hemolysis**
- * **With microspherocytosis, the minimum osmotic resistance is 0.8-0.9% NaCl**

III.2. Biochemical studies

- * Bilirubin and its fractions**
- * Total protein and its fractions**
- * M-gradient - electrophoresis of plasma proteins and urine proteins**
- * Immunoelectrophoresis - a class of immunoglobulins**
- * LDH**
- * Transaminases level**
- * Creatinine level**
- * Calcium level**

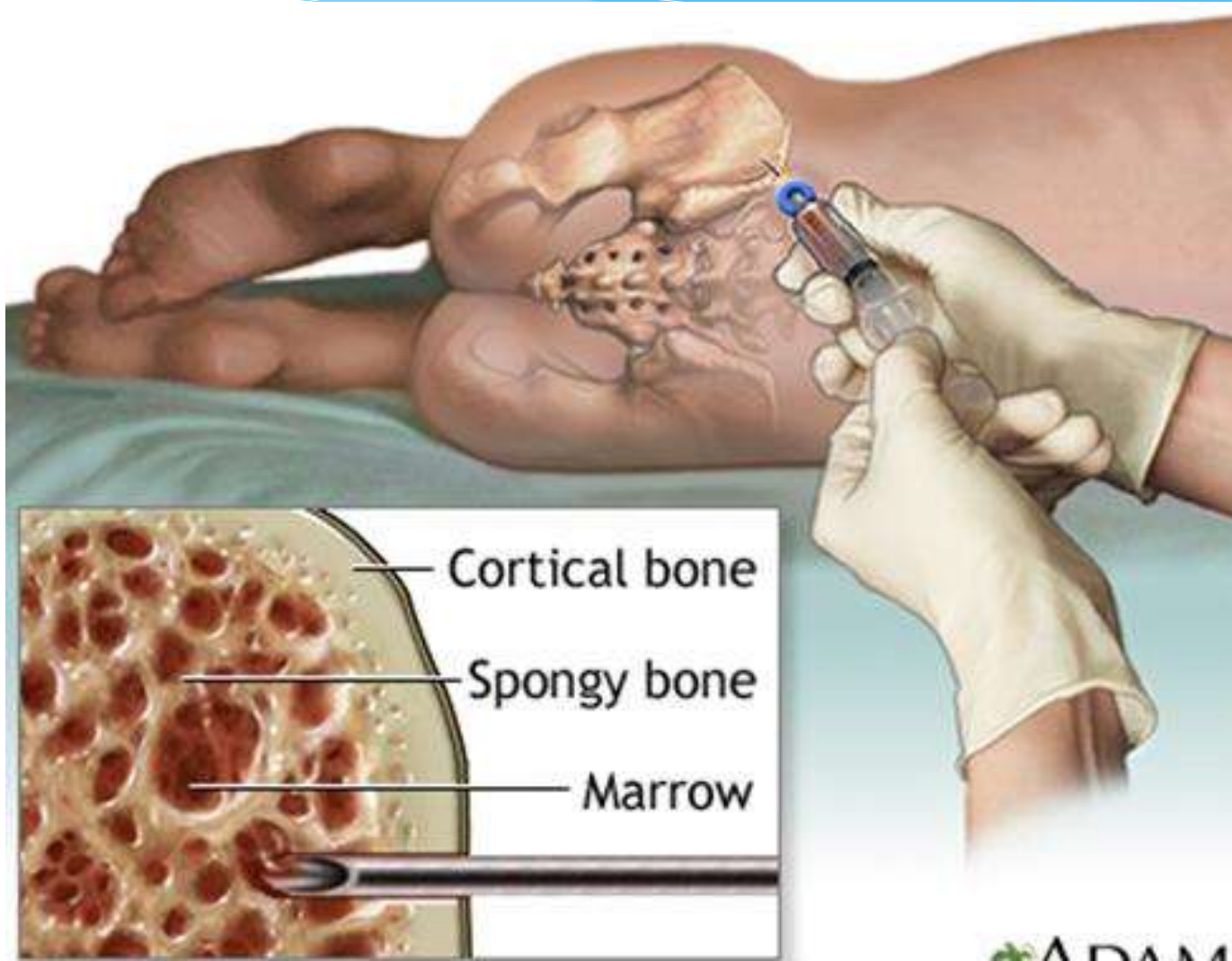
III.3. Bone marrow examination

(Sternal puncture, trepan biopsy of the iliac bone)

Sternal puncture - indications:

- *Anemia (except iron deficiency)**
- *Different cytopenia - the exclusion of hemoblastosis, aplasia)**
- *Suspected acute leukemia - diagnosis, control treatment**
- *Suspicion of chronic leukemia - diagnosis, treatment control**
- *Diagnosis of blood leukemoid reactions**
- * Increased ESR - the exception of MM**

Bone marrow aspiration and biopsy.



Bone marrow aspiration and biopsy.



Jamshedi Needle
(Click to enlarge image)



Morphological analysis of bone marrow cells

- * **Explore 500 cells**
- * **Cellularity of BM**
- * **The qualitative characteristic of all cell series**

Determination of the maturation indices

- **Neutrophils**
- **Erythroid cells**

Leukoerythroblastic ratio:

- **Granulocytes / nucleated erythroid cells = 3-4 / 1**

Determination of the hematopoiesis nature

Bone marrow

- * The number of myelokaryocytes in the norm is $50-250 \times 10^9 / l$ - reflects the cellularity of the bone marrow:

Normo-, hypo- and hypercellular marrow

Increased myelokaryocytes:

- * Acute and chronic hemoblastoses,
- * B12-deficiency anemia,
- * Hemolytic anemia
- * After acute hemorrhage.

Reduction of bone marrow cellularity:

- * Hereditary and acquired hypoplastic anemia,
- * At toxic effect of medication,
- * MDS (MyeloDysplastic Syndromes), IMF (Idiopathic MyeloFibrosis),
- * Metastases of solid tumors in the bone marrow.

Bone marrow

Megakaryocytes $0.05-0.1 \times 10^9 / L$.

↑ megakaryocytes:

- * Myeloproliferative diseases: PV, CML, IMF,**
- * immune thrombocytopenia,**
- * metastases of solid tumors in the bone marrow**

↓ megakaryocytes:

- * Aplastic anemia,**
- * Acute leukemias,**
- * Non-Hodgkin's Malignant Lymphomas**

III.4. Cytochemical study

- * Myeloperoxidase is an accessory of myeloid cells (lymphoblasts do not contain MPO) - as the cells mature, the color intensity increases.**
- * Lipids are positive in myeloid cells: promyelocytes, myelocytes, mature neutrophils, eosinophils.**

Cytochemical study

PAS (Periodic Acid–Schiff) reaction to glycogen:

- *in myeloid variants of AL - PAS-reaction is slightly diffuse or negative,**
- * with the B-cell "common" version of AL - PAS-response positive granular, with T-cell variant AL - negative**
- * α -naphthyl esterase inhibited by sodium fluoride - positive in monoblasts, monocytes**

Blast cells cytochemical study

- * **Definition of an acute leukemia variant**
- * **Determination of CML blast crisis variant**
- * **Allows you to clearly establish the type of cells, the stage of their maturation,**
- * **Determine the characteristic enzymatic reactions based on the evaluation of a specific color of different intensity:**
 - * **(-) negative reaction,**
 - * **(+) slightly positive,**
 - * **(++) positive,**
 - * **(+++) strong positive reaction.**

III.5. Immunophenotyping

- * The method for determining on the cell surface antigens differentiation of CD "differentiation clusters" (membrane and intracellular), characteristic for determining the hemopoiesis line**

The main tasks of immunophenotyping

With AML:

- * **Specification of the diagnosis: M0, M6, M7**
- * **Determination of immune prognosis factors**

With tumors of the lymphoid series

- * **Establishment of linearity of malignant cells**
- * **Determination of the stage of differentiation**
- * **Determination of the tissue equivalent of malignant tumors: NZL from cells of the mantle zone, follicular lymphomas, marginal zone lymphomas**

Determination of normal non-tumor equivalent of a malignant tumor

III.6. Immunohistochemical study

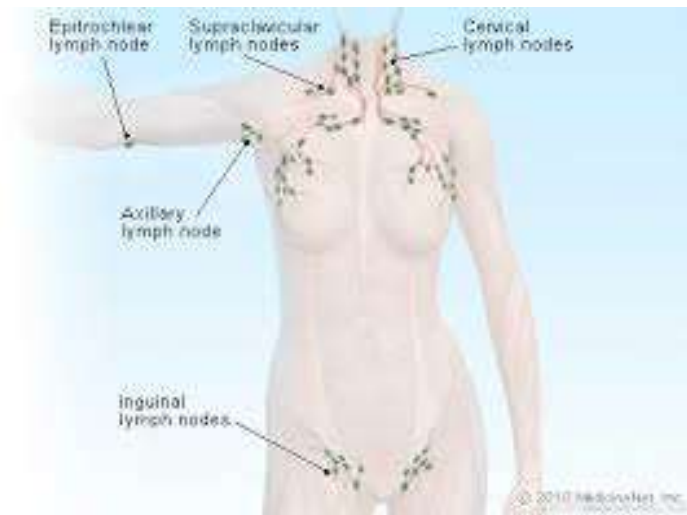
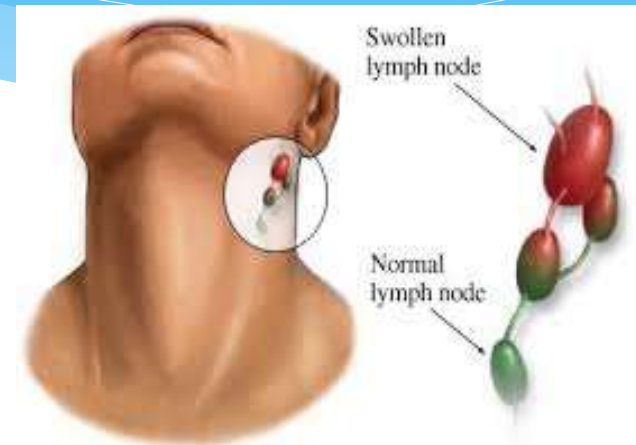
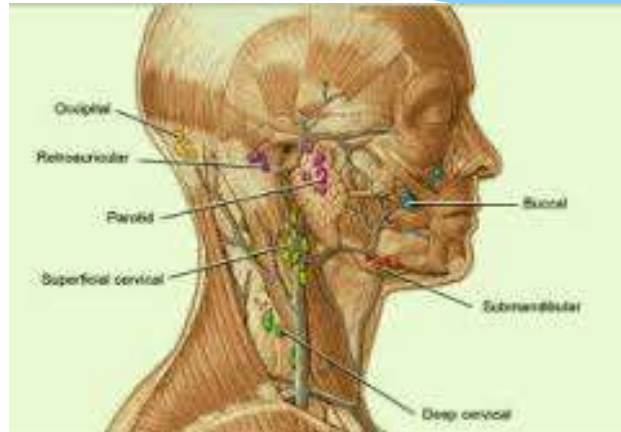
- * is a method of morphological diagnostics, based on the evaluation of antigen-antibody reactions in biopsy sections using a microscope.

In the immunohistochemical study, the following are analyzed:

- * The presence of staining,
- * Staining intensity
- * Spatial staining of cytoplasm and cell membranes, nuclei, etc.

General principles: the use of a panel of MCA responsive to CD and subsequent evaluation of the antigen-antibody in tissue biopsy sections.

Puncture of the lymph node



Possibilities of immunohistochemistry

- * **Specification of tumor histogenesis**
- * **Determination of the source of metastasis**
- * **Immunophenotyping tumors of hematopoietic and lymphoid tissue**
- * **Assessment of the functional state of the tumor**
- * **Diagnosis of immunocomplex and autoimmune diseases**

Flow cytometry

- * is performed on blood, bone marrow, body cavity fluids (peritoneal, pleural), and aspirates of solid tissues (e.g. lymph node) placed into liquid media. This is generally used for**
 - * - determination of cell lineage in lymphoma and leukemia**
 - differentiation between reactive and neoplastic expansions of lymphocytes**
 - determination of lymphocyte subsets.**

III.7. Cytogenetic study

- * **Karyotyping with the determination of the quantitative composition of chromosomes and the identification of chromosomes by structure: normal or aberrant**
- * **Chromosomes are examined in the stage of prometaphase and the metaphase of mitosis - the chromosome is maximally condensed, placed in one plane in the center of the cell separately from each other**

Chromosomal abnormalities can be:

- * *Quantitative* - karyotype with loss or addition of a chromosome
- * *Qualitative* - structural, due to restructuring, loss or addition of individual chromosomal fragment

Цитогенетичний звіт

ПІБ:

Дата

народження

Код пацієнта:

Клінічний

діагноз:

KM12125

?XML

Матеріал:

КМ

Номер скла:

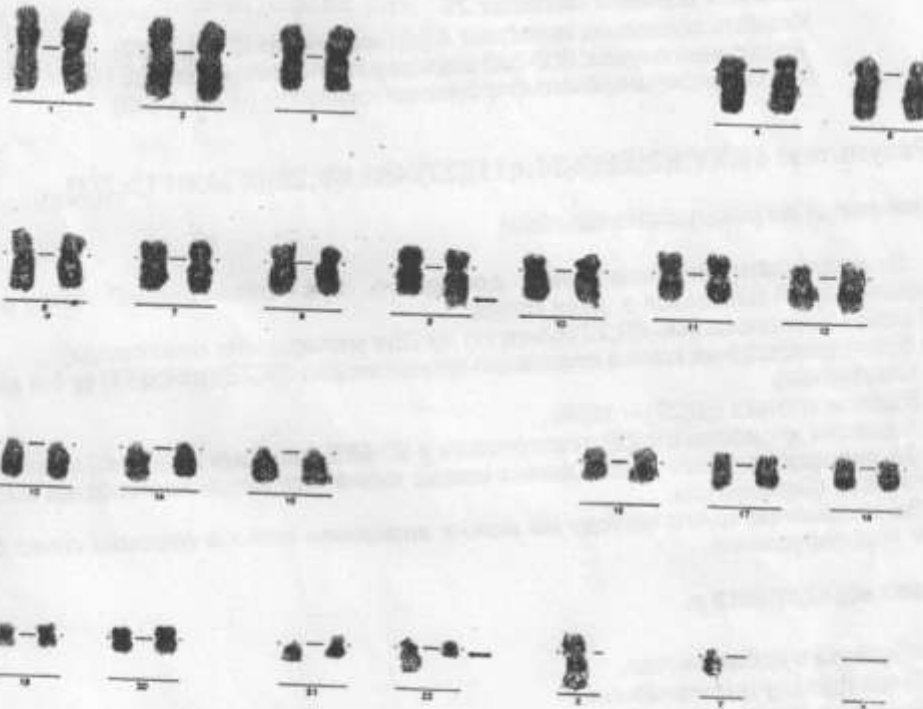
ПЗ

Пофарбування:

Координати:

GTG

143.0-8.0



Каріотип:

46,XY,t(9;22)(q34;q11)

Заключення:

Дата:

6/27/2012

Судова

Андреева С.В.

III.8. Molecular genetic research

- * **Genetic information is recorded on DNA as a linear sequence of 4 basic nucleotides: adenine, thymine, guanine and cytosine**
- * **The principle of complementarity of DNA and RNA:**
 - **Adenin - Timin**
 - **Guanine – Cytosine**

Methods of molecular genetic research:

- * **- FISH**
- * **- PCR**

Molecular genetic research

- * **FISH - (*Fluorescence In Situ Hybridization*)** is based on the hybridization of the synthetic oligonucleotide of the DNA probe to specific sequences of the studied DNA (template DNA) that are labeled with fluorochromes followed by visualization using fluorescence microscopy
- * **The sensitivity of the DNA hybridization method makes it possible to identify one tumor cell among 100-1000 cells**

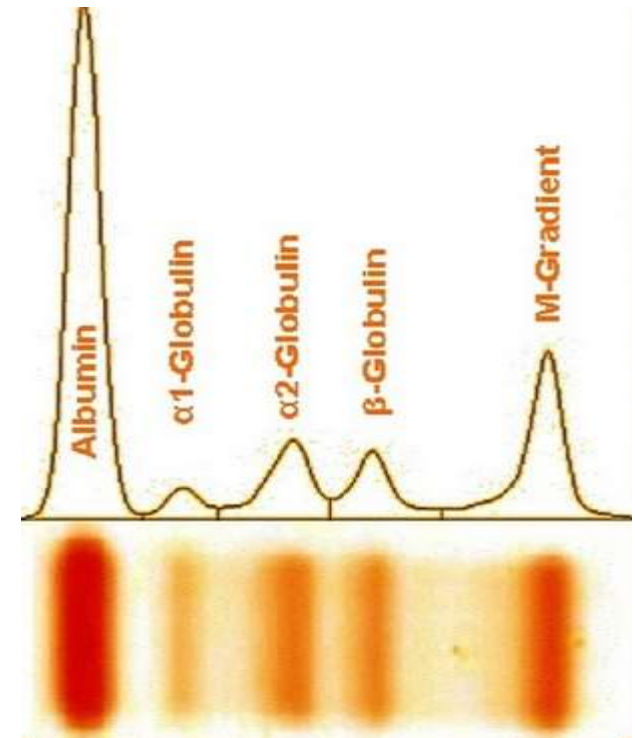
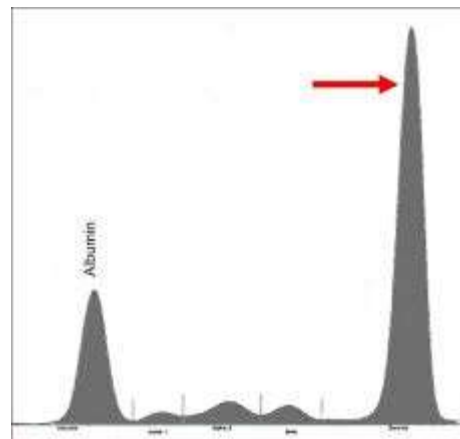
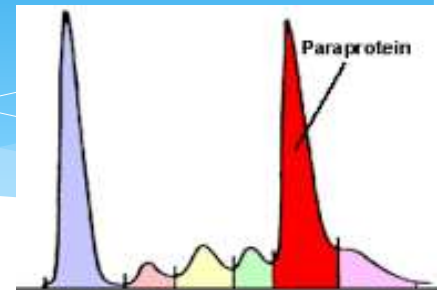
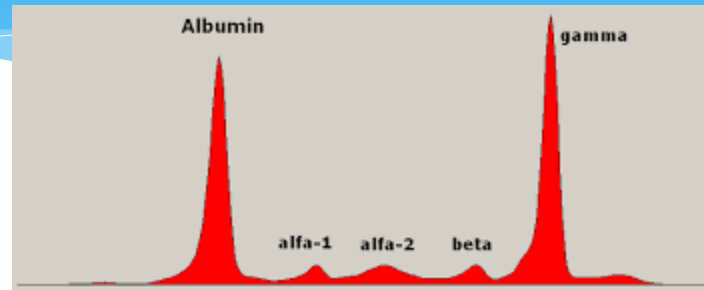
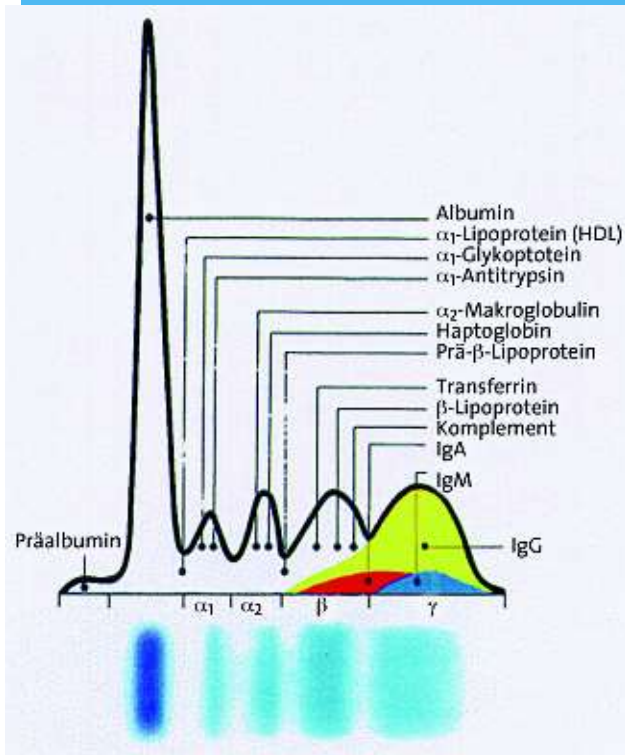
PCR:

- * Performs effective amplification (doubling in each cycle of the studied DNA molecules up to 20-30 cycles) of selected DNA sequences**
- * Has a high sensitivity - it makes it possible to identify one tumor cell by 10^5 - 10^6 normal cells**
- * Any violation of the complementarity of DNA nucleotides - mutations**

III.9. Immunochemical methods

- * **Diagnosis of immunoglobulinopathy**
- * **Electrophoretic methods - protein electrophoresis (acetate-cellulose membrane, agarose gel):**
 - * **albumins**
 - * **α 1-antitrypsin, α 1-lipoprotein- α 1-zone**
 - * **α 2-macroglobulin, haptoglobin- α 2-zone**
 - * **Transferrin, β -lipoproteids - β -zone**
 - * **Fibrinogen - Y-zone**
 - * **Immuno-fixed electrophoresis**

M-Gradient



Клинический Лабораторный Центр

Date : 07-12-2011

Prog.Nb: 25

Depart. : Foltava

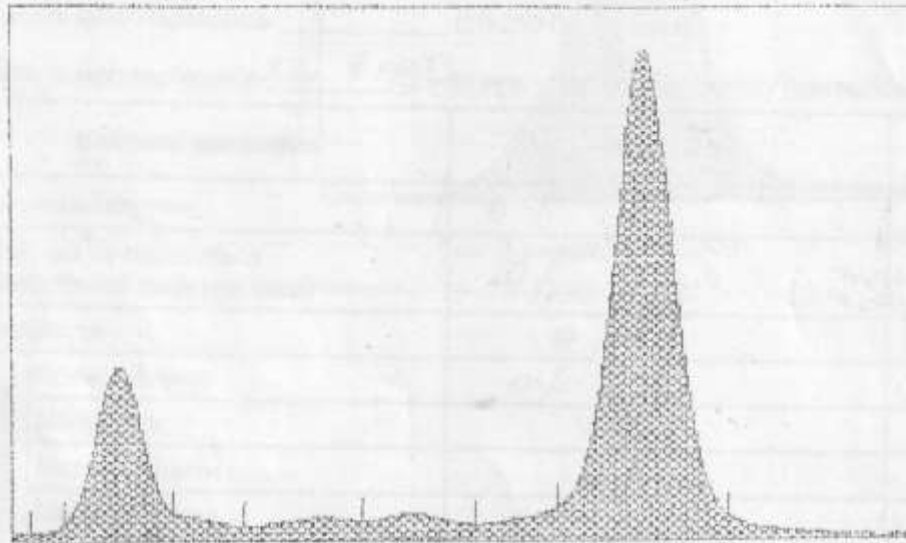
Cod.Nb :

Name : **МАНЬКО** Ser.6.12.11

Age : 45

Doctor : Ljuba

Sex : M



Name fr.	%	(G/L)	Normal val.
Proteine	0.4	0.5	0.0 - 1.0
Albumin	< 19.1	25.3	32.0 - 45.0
Alpha 1	2.2	2.9	2.0 - 4.5
Alpha 2	< 3.7	4.4	10.0 - 15.0
Beta 1	< 4.0	0.3	6.0 - 13.0
Beta 2	< 2.7	3.7	6.0 - 13.0
μ -fraction	67.7	90.2	10.0 - 20.0

Note: proteine : 13.10 (G/L) A/G : 0.4

Comment: Впрашеная диспротеинемия за счёт
наличия в этой фракции μ -глобулинов массивного
моноклона. Для уточнения типа этого моноклона
дополнительное исследование сыворотки крови мо-
носом иммунофлюоресценции.

Doct. of the lab.: Susach Natalia

Coagulopathy

- * **No single test or combination of tests is adequate to diagnose disseminated intravascular coagulation (DIC).**
- * **Perform screening tests in all patients, such as platelet count, prothrombin time (PT), activated partial thromboplastin time (aPTT), thrombin time, and fibrin degradation products or soluble fibrin monomers.**
- * **Additional tests that may be useful include antithrombin III levels, protein C, D-dimer, fibrinogen, measurements of specific coagulation factors such as factor V and VIII, and plasminogen activator inhibitor type I (PAI-1).**
- * **Clinical judgment in conjunction with these tests provide a means of working towards a diagnosis of DIC, although no single test results alone are confirmatory.**

Coagulopathy

- * Thrombocytopenia is an almost universal finding, and the CBC count with smear review may reveal findings suggestive of DIC, such as increased platelet size, schistocytes and helmet cells.**

Coagulation Parameters

- * **Prothrombin time (PT)** **11-14 seconds**
- * **Partial thromboplastin time (PTT)** **25-35 seconds**
- * **International normalized ratio (INR)** **0.8 to 1.2**

Methods of radiation diagnostics

- * **X-ray methods**
- * **CT scan**
- * **Magnetic Resonance Tomography**
- * **Radionuclide methods: scintigraphy, positron emission tomography**
- * **Ultrasonographic methods**

*Multiple myeloma pathognomonic
symptom (bone lesions)*



**Thank you
for your attention!**

